

A SCANNING ELECTRON MICROSCOPIC INVESTIGATION OF THE POLLEN OF SOUTH AFRICAN *ACACIA* SPECIES

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ABSTRACT

Pollen of South African *Acacia* species was used as experimental material for studying techniques of scanning electron microscopy. Additional information on the pollen morphology of *A. albida* and *A. giraffae* was thus obtained. The taxonomic position of *A. albida* and *A. giraffae* is discussed briefly.

Microsporogenesis of *A. exuvialis* was examined as well as the surface structure of the anthers and their glandular appendages.

UITTREKSEL

'N SKANDEERELEKTRONMIKROSKOOP ONDERSOEK VAN DIE STUIFMEEL VAN SUID-AFRIKAANSE *ACACIA*-SOORTE.

Stuifmeel van Suid-Afrikaanse *Acacia*-spesies is gebruik as eksperimentele materiaal om tegnieke op die skandeerelektronmikroskoop te bestudeer. Hierdeur is bykomstige inligting in verband met die stuifmeelmorfologie van *A. albida* en *A. giraffae* verkry. Die taksonomiese posisie van *A. albida* en *A. giraffae* word kortliks bespreek.

Die mikrosporogenese van *A. exuvialis* sowel as die oppervlaktestruktuur van die helmknoppe en die klieragtige aanhangsel van die helmknop is ondersoek.

INTRODUCTION

Acacia pollen has previously been studied by Rosanoff (1866), Wodehouse (1935), Erdtman (1953, 1952), Selling (1947), Coetzee (1955), van Zinderen Bakker and Coetzee (1959) and Guinet (1969).

The three dimensional structure of the polyads and monads are clearly illustrated in these papers, but very little detail of the surface structure of the cells is shown. Van Zinderen Bakker & Coetzee (1959) have indicated that all the polyads of all South African *Acacia* species consist of 16 cells (monads) with the exception of *A. albida* (with 30 cells) and *A. giraffae* (with 26-48 cells). Coetzee (1955) has also mentioned that the monads of species with spicate inflorescences as well as those of *A. detinens* (*A. mellifera* var. *detinens*) and *A. pennata* (*A. schweinfurthii* pro parte) (with capitulum-like inflorescences) are not furrowed (grooved), whereas species with capitate inflorescences have grooved monads. According to characteristics other than pollen structure, *A. mellifera* and *A. schweinfurthii* belong to the series *Vulgares* (Benth., 1875). Guinet (1969) therefore correctly states that species of the series *Vulgares* (with

spicate inflorescences, including *A. mellifera* and *A. schweinfurthii*) have un-grooved monads, while species of the series Gummiferae (with capitate inflorescences), except *A. albida*, have grooved monads.

According to Coetzee (1955), the furrows (grooves) in the monads of *A. giraffae* are the of same type as those in monads of the remaining species with capitate inflorescences. Guinet (1969), by contrast, shows clearly that the grooves in the monads of *A. giraffae* are different from those in the remaining species of the Gummiferae. Neither Coetzee (1955) nor Guinet (1969), however, gives a detailed illustration or description of the polyads of *A. giraffae*.

In view of the large amount of available information on *Acacia* pollen, this study was not intended to be a complete revision of the work that has already been done on *Acacia* pollen. The pollen of some South African *Acacia* species was used merely as experimental material for studying techniques of scanning electron microscopy (SEM). A comparison of the observations made during this study, with observations on *Acacia* pollen made previously, was nevertheless considered relevant.

As it was not easy to distinguish the three dimensional structure of grooved monads with the SEM, it was necessary to compare scanning electron micrographs of the polyads with light-micrographs.

In order to understand the ontogeny of the polyads better, the microsporogenesis of *A. exuvialis* was also examined with attention being paid to the structure of the anther and its grandular appendage.

MATERIALS AND METHODS

Fresh pollen and/or pollen from herbarium specimens (untreated) was mounted on specimen holders, coated with gold palladium or gold and then studied on a Jeol JSM-U3 scanning electron microscope. Pollen of *A. albida*, *A. giraffae* and *A. exuvialis* was pretreated in a solution of 1 ml H_2SO_4 and 9 ml acetic anhydride (Erdtman, 1943) and studied under the light microscope as well as the SEM.

In order to study the microsporogenesis of *A. exuvialis*, inflorescences at different stages of development were fixed in glutaraldehyde and embedded in glycol methacrylate. Sections of 1–2 μm were cut using an ultramicrotome and then stained with toluidine blue.

RESULTS AND DISCUSSION

A. Microsporogenesis

A. exuvialis produces flowers over a prolonged period of time (October to April) and inflorescences at different stages of development can be found on the same tree. Fig. 1 shows a very young anther in which the sporogenous tissue has not yet formed. Fig. 2 shows the dividing microspore mother cells. Eight microspore

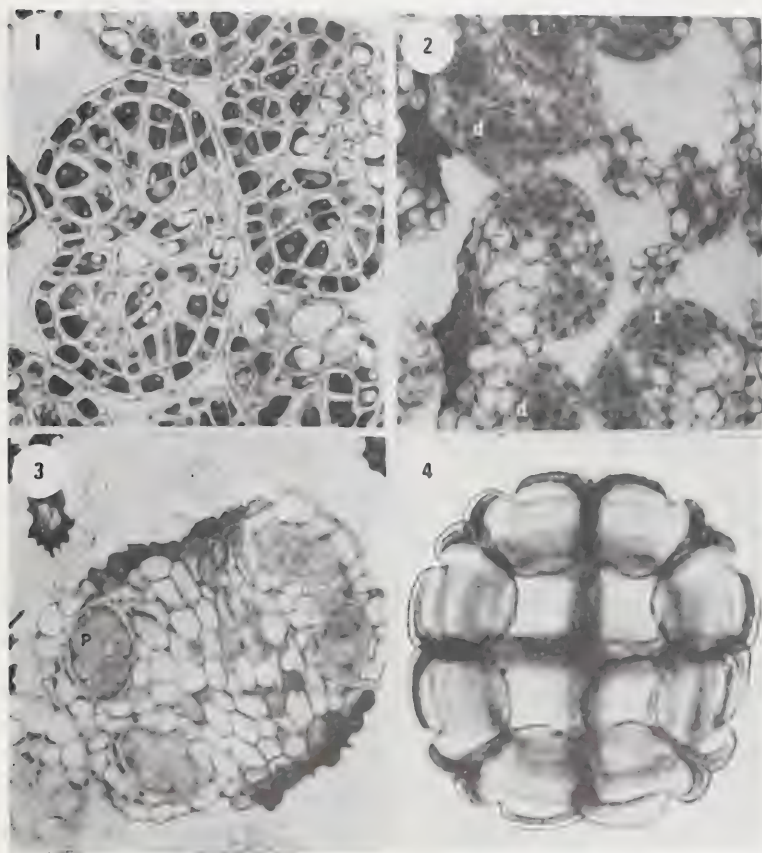


FIG. 1-3.

Part of a section through a young inflorescence of *Acacia exuvalis* showing: 1, very young anthers; 2, microspore mother cells after the first meiotic division of the nucleus, and tetrads; 3, polyads in the anther lobe.

d = diad; p = polyad; t = tetrad.

FIG. 4.

A light micrograph of a polyad of *A. exuvalis*.

mother cells are formed in each anther—four in each lobe. The four mother cells in a lobe are separated transversely by sterile cells, unlike the continuous row of pollen mother cells in each of the two cylindrical locules of the anther lobe found in most other Dicotyledonous plants (Maheshwari, 1950). After meiosis, one tetrad is formed from each microspore mother cell (Fig. 2). Subsequent mitotic divisions within each tetrad then give rise to a 16 cell polyad so that, ultimately, eight polyads are formed in each anther (Fig. 3).

Acacia exuvialis is a member of the series Gummiferae and, as expected, the polyads of this species are grooved. The centrally situated monads have a Y-shaped groove with two outer ridges, while the peripherally situated monads have H-shaped grooves with three peripheral ridges like those of most other Gummiferae species (Fig. 5B).

B. The Anther

Maheshwari (1950) states that the greater part of the mature anther wall consists of the subepidermal layer (endothecium) of the anther wall, since the epidermis, as well as the inner cell layers, becomes crushed as the anther matures. He also states that, in these subepidermal cells, fibrous bands (ridges) stretching as far as the outer tangential walls are formed on the radial walls.

Scanning electron micrographs of the mature anther wall show the remains of the epidermal cells and the corrugated outer walls of the endothecial cells (Fig. 6C). These corrugations are caused by the mentioned ridges stretching over the tangential walls as well.

The anthers of all South African *Acacia* species except *A. albida* have glandular appendages. Figure 6D shows a scanning electron micrograph of a young anther gland of *A. kirkii* with the secretion between the cells. In older glands the secretion has dried up and the cells appear very prominent.

C. The Polyads

Prior to this study, *Acacia* pollen has been studied largely under the light microscope which shows clearly the cellular structure of the polyads (Fig. 4). Little detail of the surface structure of the polyads, however, is visible.

With the aid of the scanning electron microscope much greater detail of the surface structure of the polyads can be seen.

Polyads with ungrooved monads:

Work with the scanning electron microscope confirms previous reports that the polyads of all the South African *Acacia* species which belong to the series Vulgares, as well as *A. albida*, have ungrooved monads. With the exception of *A. albida*, all of these polyads consist of 16 cells (Fig. 5A).

According to Van Zinderen Bakker and Coetzee (1959) there are 30 monads per polyad in *A. albida* and according to Guinet (1969) there are 16–30, but most

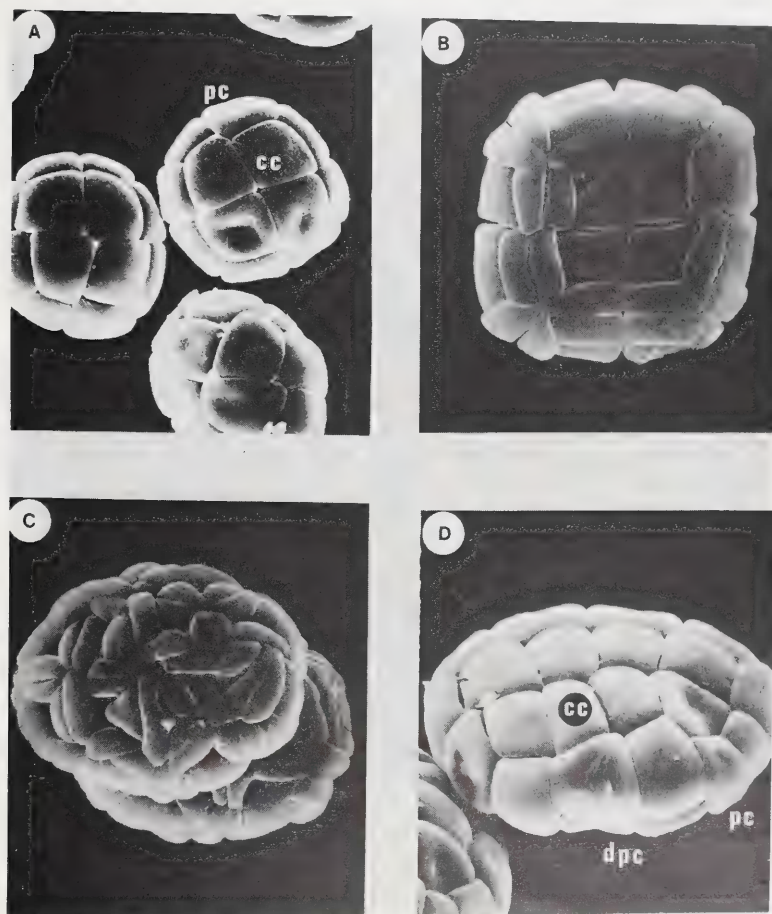


FIG. 5.

Scanning electron micrographs of polyads of

A. *Acacia hereroensis*, B. *A. borleae*, C. *A. giraffae* and D. *A. albida*.

cc = central cells; pc = peripheral cells; dpc = double layer of peripheral cells.

frequently 30. From Figure 5D it is evident that polyads can also consist of 32 cells. *A. albida* differs from all the other South African *Acacia* species in that two peripheral monads on each lateral side of a polyad undergo a single division to form a double layer, while the rest of the peripheral cells remain single. In the polyads of all other *Acacia* species (except *A. giraffae*) the peripheral cells occur in a single layer.

Polyads with grooved monads:

In pollen treated with acid, individual cells in the polyad are easily identifiable under the light microscope (Fig. 4). However, when the SEM is used, the grooves are so prominent that the ridges can easily be mistaken for individual cells (Fig. 5B).

This study further confirms previous observations that polyads of the South African acacias which belong to the series Gummiferae (except *A. giraffae*) consist of 16 cells, all of which are grooved (Fig. 5B, 6A and 6B). As in the case of the polyads of the Vulgares, the 16-celled polyad comprises a double layer of central cells (eight in total), surrounded by 8 peripheral cells. The polyads are usually very uniform in shape and extremely symmetrical. The surface structure is not very conspicuous but does show indentations (Fig. 6A and 6B).

According to Coetzee (1955) polyads of *A. giraffae* consist of 32 cells. Van Zinderen Bakker and Coetzee (1959) give the number of monads as 26–48 and Guinet (1969) as 16–32. Apart from the difference in the number of monads per polyad it is quite clear from Fig. 5C that the polyads of *A. giraffae* are different from those of other Gummiferae species. Without the aid of the light microscope it is not at all easy to distinguish the number of monads in scanning electron micrographs of *A. giraffae* polyads. Once the configuration of the cells has been determined (by light microscopy) the shapes of the grooved cells can easily be recognised under the SEM. In polyads of *A. giraffae* the monads are not clearly arranged in centrally- and peripherally-placed cells as in the other species of Gummiferae. The number of monads per polyad varies from 26–48. The monads are irregular in shape and have grooves varying from Y-shaped and H-shaped to X-shaped (Fig. 7). The ridges forming these grooves have a granular appearance.

DISCUSSION

Chevalier (1934) considered *A. albida* sufficiently different from other *Acacia* species to be placed in the monotypic genus *Faidherbia*. This new generic name has been retained by Guinet (1964 and 1969) on the basis of the anomalous pollen morphology of *A. albida*. Wickens (1969), too, has commented on the taxonomic position of *A. albida*, while Robbertse (in press) groups this species, together with *A. schweinfurthii*, *A. brevispica* and *A. kraussii* in “a some-

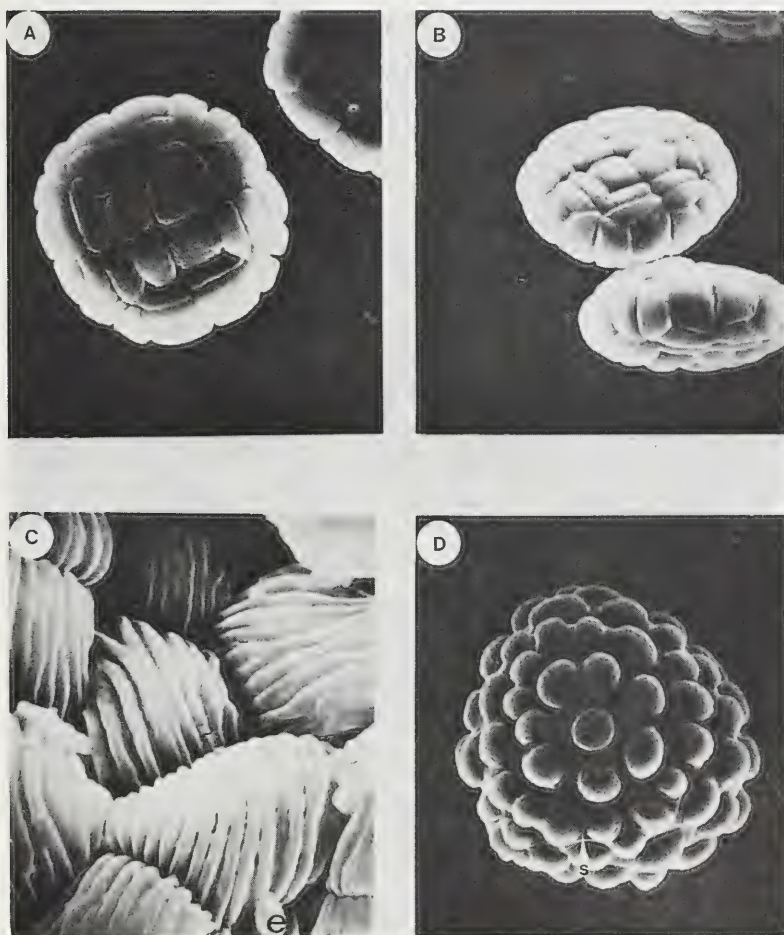


FIG. 6.

Scanning electron micrographs of polyads (A + B) and anther gland (D) of *A. kirkii* and a surface view of the endothelial cells from the anther of *A. giraffae* (C).
 e = remains of epidermal cell; s = secretion.

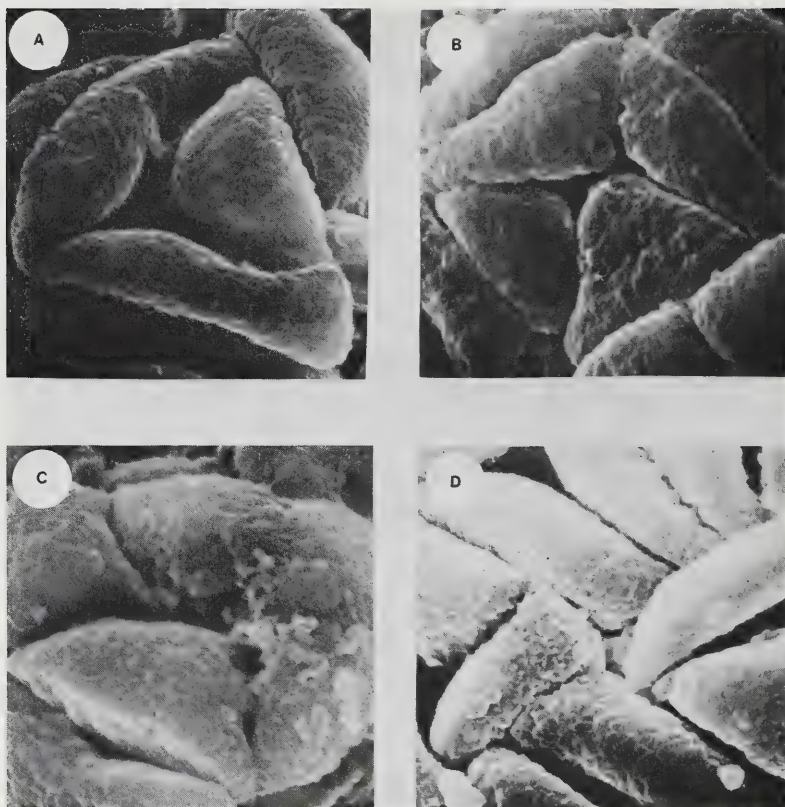


FIG. 7.

Scanning electron micrographs of monads from a polyad of *A. giraffae*, showing the structure of the monads.

A—monad with Y-shaped groove. B & C—monads with H-shaped grooves. D—granular structure of ridges.

what artificial" series (subgenus), *Farinosae*, on the basis of seed, seedling and flower morphology.

The degree of difference between the polyads of *A. albida* and those of the *Vulgares* species is, however, not greater than that between the polyads of *A. giraffae* and those of the rest of the *Gummiferae* species. For this reason and on the basis of other morphological criteria *A. albida* and *A. giraffae* must be seen as possible relicts of the parental stock of the remaining African *Acacia*

species rather than regarding *A. albida* as representing a completely distinct genus.

Scanning electron microscopy, as a technique, is a valuable aid to the plant morphologist. It should be regarded as an important supplement to light microscopy but cannot be used alone as a substitute.

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